by Kepecs et al. (2008), they demonstrated that the activity of neurons in the rat orbitofrontal cortex (OFC) matched the model of the rat's uncertainty regarding their own past decision. Metacognitive signals in the corresponding area in monkeys should thus be examined in future studies, which will facilitate our understanding of the relationships between the metacognitive signals in different brain areas (Figure 1C).

The strength of the metacognitive signal observed in Middlebrooks and Sommer (2012) was several spikes per second on average, which is not a large proportion of all the spikes fired by these neurons. Therefore, readout mechanisms and the behavioral impact of the observed metacognitive signals should be considered carefully. This is related to the issue of across-areal neuronal circuitry for metacognition, which would include the SEF, LIP, and presumably OFC, among which anatomical connections have been identified (Figure 1C) (Cavada et al., 2000; Lynch and Tian, 2006). Clarifying the hierarchical relationships between these areas and differentiating their roles in metacognition should be the next step in understanding the neuronal circuitry that implements this cognitive process, which we humans profoundly exploit to lead our daily lives.

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Losing the Lust for Life: A New Role for an Old Feeding Peptide?

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A recent paper in *Nature* (Lim et al., 2012) describes the effects of melanocortin receptors in the nucleus accumbens. The studies connect a hypothalamic peptide system with brain reward centers and show effects on specific neuronal populations and behavioral components of mood.

Food and Mood

It is hard to imagine something more integrated with our mood state than eating. The influences go in both directions, with intake affecting mood and mood states modulating eating. For example, depression can lead to either increases or decreases in intake. As with all complex neuropsychiatric conditions, elucidation of basic neurobiological mechanisms is a critical first step toward clarifying just how the brain integrates eating with emotions. A recent study from Robert Malenka and colleagues published in *Nature* identifies molecules, circuits, and neuronal pathways by which hypothalamic derived peptides can influence hedonic states (Lim et al., 2012). Specifically, the study establishes mechanisms by which stress can lead to reduced intake and anhedonia.

Melanocortins and Their Receptors—Taking a Hint from Metabolism

The melanocortin agonist, alpha-MSH, is derived from the precursor peptide POMC. The POMC neurons of the arcuate nucleus form the "stop" side of the hypothalamic feeding equation whereby activation of this population reduces intake. The paraventricular nucleus of the hypothalamus has been best studied as a site where the melanocortin MC4 receptor (MC4R) mediates these effects. However, the MC4R is broadly expressed in the brain, including the nucleus accumbens and dorsal striatum. Early work showed regulation of MC4R by opiates and a role for striatal MC4R signaling in cocaine reward (Alvaro et al., 2003; Hsu et al., 2005), and more recent studies have

shown that MC4R is present on dopamine receptor-1 (D1)-expressing medium spiny neurons that are needed for procedural leaning (Cui et al., 2012). Previous findings implicate MC4R in stress responses and anxiety but did not identify brain regions involved (Chaki and Okuyama, 2005). Now, Lim et al. (2012) integrate this previous work and add a wealth of new mechanistic and behavioral data. They start by establishing that POMC neurons project from the arcuate nucleus to the core region of the nucleus accumbens. This mapping sets the anatomical stage for a more detailed neuronal and functional analysis.

Synaptic Specificity and Long-Term Plasticity

Through brain-slice electrophysiology studies, the authors find similar effects of alpha-MSH and stress on medium spiny neurons (MSNs) of the nucleus accumbens. Both reduce excitatory postsynaptic currents (EPSCs) via alterations of AMPA receptor subunit composition, as supported by observed changes in rectification. Strikingly, the effects of stress and MC4R agonism are only apparent on D1 neurons, whereas neither affects D2 neurons. Moreover, the effects of stress appear to depend on MC4R signaling in the region, which is significant because MC4R protein is upregulated during stress. Together, the findings support a physiological role for changes in MC4R signaling during stress-induced adaptation in the region.

The changes in synaptic strength were then examined for effects on long-term depression (LTD). Pre-exposure to alpha-MSH occluded LTD, and this effect is shown to depend upon MC4R. This LTD appears also to be AMPAR subunit-dependent since it is sensitive to treatment with NASPM. To better relate the LTD to AMPA receptor dynamics, the authors used a virus expressing G2CT-pep, a synthetic peptide designed to prevent internalization of Glua2 expressing AMPARs. This in vivo manipulation caused a reduction in LTD while also blocking behavioral responses to stress.

With the effects of MC4R on synaptic and neuronal signaling characterized, the authors asked how MC4R could have these effects on D1 neurons. Gs signaling through D1 receptors has been very well studied and primarily occurs via effects on cAMP-dependent protein kinase A (PKA). Since MC4R is also Gs coupled, it is not clear how its effects could be distinguished from D1 signaling. While there are many details to work out, the paper provides a first clue by identifying the alternate EPAC2 (cAMP-activated postsynaptic protein) as a critical part of the signaling that affects stress responses.

Anhedonic States

So what are the consequences of MC4R signaling on animal behavior and mood? Stress-induced weight loss is the behavioral assessment used for most of the experiments. The mice lose weight during 8 days of restraint stress, which is accompanied by reduced food intake. The authors interpret this as a stress-induced anhedonia and then find support for this with sucrose preference, which is also reduced by stress via MC4R signaling in the accumbens core. These effects of stress are blocked when MC4R receptor levels are reduced using shRNA. Of course, traditional gene knockdown using shRNA affects all neurons, so the possibility of an indirect effect of reduction of MC4R in D2 MSNs or other neurons is possible. To address this, the authors used a creative viral approach that utilized Cre recombinase to selectively re-express an shRNA resistant MC4R in D1 neurons of the nucleus accumbens. These animals had a normal stress response, confirming that MC4R function in D1 neurons of the accumbens is sufficient to produce anhedonia.

Strikingly, other measures of antidepressant efficacy, the forced-swim and tail suspension tests, were not affected by either MC4R gene knockdown or G2CT-pep administration in the nucleus accumbens. These tests are mainly used for their predictive validity but are also thought to represent behavioral despair in animals. The effects of MC4R on sucrose preference and food intake are perhaps not surprising given MC4R's general role in ingestive behavior. In fact, the reliance on intake as a measure of hedonic response can be problematic since it can be modified by metabolic state. However, a more general role in reward was revealed in the final experiments, where MC4R is shown to be essential for the reduction in cocaine place preference in response to stress. That stress reduces place preference is noteworthy given that in other models of stress and reward, stress increases drug seeking in both place preference and reinstatement tests (Bruchas et al., 2010). However, these stressors tend to be more acute, and a persistent, chronic stress used here is likely responsible for the opposing results.

There remains a question of how these findings might relate to the constellation of behaviors underlying depression, and here we face the problem of modeling a complex disease in animals. In this case, it will be interesting to look at other elements of depression, including anxiety and social defeat stress. These models would help to connect MC4R to previous studies in the nucleus accumbens that have shown reduced activity of medium spiny neurons leading to greater anxiety and susceptibility to social defeat (Wallace et al., 2009).

Another important question that arises from this study is how this pathway is selectively activated during stressful conditions since the MC4R effects interact with stress. The answer likely lies in the regulation of both alpha-MSH and the MC4R. Alpha-MSH production and release are specifically upregulated due to emotional stress (Liu et al., 2007), while the current studies demonstrate that MC4R levels are increased in a linear fashion with increasing days of stress. So this selective tuning of both agonist and receptor is able to rapidly convey changes in physiological signals to a specific neuronal population.

Getting to the Core of the Problem

The authors' targeting of nucleus accumbens core is significant because research looking into ventral striatal control of food intake has segregated the function of the core and shell. Opioid induced intake of highly palatable food is broadly controlled across the shell and core (Baldo and Kelley, 2007). In contrast, glutamate antagonist- or GABA agonist-induced intake of normal chow is found only within the medial aspect of the shell. It remains to be seen if similar effects on MC4R activity can influence food intake if targeted in the shell

subregion. Since MC4R is inhibiting neurons, it is possible that the effects would not be as robust in the shell, where inhibition would be predicted to lead to increased intake. Finally, existing literature on the nucleus accumbens and hedonic state has focused on the shell region (Barrot et al., 2002), making the current work unique in its emphasis on the core.

The present work also adds to the list of characteristics that distinguish D1 and D2 neurons. While many obesity studies have focused in D2 neurons, the results add to evidence that D1 neurons play an important role. With D2 neurons, the model of "reward deficiency" driving intake has been proposed with some supporting animal data (Johnson and Kenny, 2010). With D1 neurons, a more traditional reinforcement model is assumed, whereby D1 stimulation leads to increased motivation to eat. However, the role of dopamine in eating and motivation is complex (Baldo and Kelley, 2007) and D1 neurons are likely to influence many components of behavior related to food intake, including the effects of stress.

From Metabolism to Affect

Hypothalamic peptides, such as MCH, have been shown to influence feeding and mood by action in the accumbens (Georgescu et al., 2005; Sears et al., 2010). The current work brings a new, important hypothalamic input into the ventral striatum. Moreover, Lim et al. (2012) provide detailed neuronal analysis as well as behavioral studies that place the circuit within the context of stress response. This combination of mechanistic analysis as well as behavioral studies make this work a major contribution to feeding, stress, and depression research fields.

As with many signaling pathways, the melanocortin pathway has been co-opted to function in different contexts. First identified as a factor that controls pigmentation in fish, melanocortin receptors also serve to control hair color in mammals. In the brain, the best-studied function is regulation of food intake and metabolism. The results from Lim et al. (2012) also implicate melanocortins in a tightly regulated stress response where they adversely affect reward and hedonic state that is relevant to depression. We know that depression presents itself in many ways, with patients suffering from different symptoms. The parsing of anhedonia and helplessness is therefore critical, and the present work gives specific mechanisms and potentially distinct targets for future therapies.

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